DATA EVALUATION RECORD

1. **CHEMICAL:** DCPA.
   Shaughnessey Number: 078701.

2. **TEST MATERIAL:** Technical DCPA; dimethyl 2,3,5,6-tetrachloroterephthalate; Code No. T-170-1; 96.7% active ingredient; a white powder.

3. **STUDY TYPE:** Freshwater fish 96-hour acute toxicity test. Species Tested: Rainbow trout (Salmo gairdneri).

4. **CITATION:** Shults, S.K. and N.H. Wilson. 1989. Static, Acute Toxicity (LC_{50}) Study in Rainbow trout with Technical DCPA (Dimethyl 2,3,5,6-Tetrachloroterephthalate). Prepared by Huntingdon Research Centre plc, Huntingdon, Cambridgeshire, England (in association with SDS Biotech Corporation, Painesville, Ohio). Document No. 690-5TX-84-0046-002. Submitted by Fermenta Plant Protection Company, Mentor, Ohio. EPA MRID Number 410548-26.

5. **REVIEWED BY:**

Kimberly Rhodes
Associate Scientist
KBN Engineering and
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Signature: Kimberly Rhodes
Date: July 30, 1990
Cheryl Sears 9/19/90

6. **APPROVED BY:**

Pim Kosalwat, Ph.D.
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KBN Engineering and
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Signature: P. Kosalwat
Date: July 30, 1990

Henry T. Craven, M.S.
Supervisor, EEB/HED
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Signature: Henry T. Craven
Date: 9/24/90

7. **CONCLUSIONS:** This study appears scientifically sound but does not fulfill the guideline requirements for an acute toxicity study using freshwater fish. The only dosage tested was <100 mg/L but not high enough to produce any effects on the test fish. The 96-hour LC_{50} value of DCPA for rainbow trout, based on the weighted mean average of DCPA was determined to be >6.6 mg/L.

8. **RECOMMENDATIONS:** The test should be repeated using a flow-through system with higher test concentrations and lower water hardness.

9. **BACKGROUND:**

10. **DISCUSSION OF INDIVIDUAL TESTS:** N/A.

11. **MATERIALS AND METHODS:**

A. **Test Animals:** Rainbow trout (Salmo gairdneri) used in the test were obtained from Parkwood Trout Farm, Medway, Kent, U.K. The fish were acclimated for 10 days at a temperature of 14 ± 1°C. The fish were fed trout pellets 5-7 times per week except during the 24-hour period prior to test initiation. No medication was administered during holding and no mortality occurred. The rainbow trout used for this study had a mean weight of 2.95 grams (g) with a standard deviation of 0.58 and a mean standard length of 5.45 centimeters (cm) with a standard deviation of 0.47. The organism loading was 0.6 g of bodyweight per liter (L) of test solution.

B. **Test System:** The test was conducted in glass aquaria containing 50 L of test solution at a depth of 27 cm. The temperature was maintained at 15 ± 1°C. A 16-hour light and 8-hour dark photoperiod was provided daily. No aeration was provided during the test.

The dilution water used for the test was dechlorinated laboratory tap water characterized as having a pH range of 7.7 to 7.8, a conductivity range of 770 to 820 µmhos/cm, an alkalinity range of 170 to 187 mg/L as $CaCO_3$, and a total hardness range of 326 to 344 mg/L $CaCO_3$.

C. **Dosage:** 96-hour static acute test. The nominal concentration was 10 mg/L.

D. **Design:** Based on the results of a preliminary range-finding study, one concentration of DCPA, a control, and solvent control were tested. A 1% solution of Tween 80 in dimethylformamide was utilized as a solvent for the preparation of stock solution of test material and for the solvent control.

The test was initiated when 10 rainbow trout were distributed to each aquarium containing prepared solution (30 fish per concentration or control). All fish were observed at 0, 3, 6, 24, 48, 72, and 96 hours for mortality and abnormal effects. Dissolved oxygen concentration, pH, and temperature were measured once daily in each replicate of the treatment level and the controls. Analytical determination of DCPA was performed from each replicate of the treatment level and in one replicate of the solvent control at each 24-hour interval using gas chromatography.

E. **Statistics:** No statistical analysis was reported as being conducted by the author.

12. **REPORTED RESULTS:** The measured test concentrations during the 96 hour test are presented in Appendix C (attached). Analysis of the test solutions showed a decline in mean measured concentration from 10.0 mg/L at the initiation of the study to 4.7 mg/L at 96 hours. The authors stated because the study was conducted at a nominal concentration well in excess of the water solubility of DCPA, it is probable that the decrease in mean measured concentration from initiation to 96 hours was partly due to the settling of the test material in the aquaria. The concentrations of DCPA in the water exceeded the level of water solubility of the material by a factor of approximately 14 at 24 hours and by a factor of approximately 9 at 96 hours.

Cumulative mortality data for rainbow trout exposed for 96 hours to DCPA are shown in Table 1 (attached). No mortalities resulted from the administration of technical DCPA to 30 trout at a nominal concentration of 10 mg/L. No abnormal signs were noted in any of the trout during this study. Since the test fish were exposed throughout the study to a mean measured concentration of DCPA technical which was greater than or equal to the concentration detected at 96 hours, the LC_{50} for the material based on the mean measured concentrations would be considered to be greater than 4.7 mg/L, the concentration detected at 96 hours.

During the study, the pH ranged from 7.8 to 8.0, the dissolved oxygen concentration ranged from 7.4 to 9.2 mg/L, and the temperature ranged from 14.2 to 14.6°C.

13. **STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:** The 96-hour LC_{50} for DCPA technical was determined to be >10 mg/L based on nominal concentration or >4.7 mg/L based on the 96-hour mean measured concentration. These values are well in excess of the level of water solubility for DCPA.

A Compliance Statement was included in the report indicating that the study was conducted in accordance with the FIFRA Good Laboratory Practice Standards set forth in 40 CFR Part 160. However, a statement from the registrant states that this study does not meet the requirements of 40 CFR Part 160 because the Compliance Statement lacks some of the required signatures.

14. **REVIEWER'S DISCUSSION AND INTERPRETATION OF STUDY RESULTS:**

A. **Test Procedure:** The test procedures were generally in accordance with protocols recommended by the SEP except for the following deviations:

o The SEP states recommends that fish be acclimated to study conditions for at least two weeks prior to testing. The fish used in this test were acclimated to study conditions for only 10 days.

o The dilution water used during this study was dechlorinated water. The SEP states that dechlorinated water should not be used as dilution water because removal of chlorine is rarely complete and residual chlorine can be quite toxic to aquatic organisms. Furthermore, this study did not measure the residual chlorine in the dilution water. The ASTM states that the concentration of residual chlorine should be <3 μg/L in fresh samples of the dilution water. However, the dechlorinated water used in this test is considered acceptable since no mortality occurred in both controls.

o A soft water with a hardness of 40 to 48 mg/L as $CaCO_3$ is recommended; this test used very hard water with a hardness of 326 to 344 mg/L as $CaCO_3$.

o The SEP states that static acute tests are initiated either by adding the test material to the test chambers after the fish are added or by adding the fish to the test chambers within 30 minutes after the test material is added to the dilution water. During this test, the time interval between the preparation of the test

solution and the addition of the fish was not specified.

o The solvent used was 1% solution of Tween 80 in DMF; Tween 80 is not a recommended solvent.

o The concentration selected for the test was less than 100 mg/L but not high enough to produce any effect on the fish.

o The SEP states that the recommended temperature for testing coldwater fish species is 12 $\pm$ 1°C. During acclimation, the temperature was 14 $\pm$ 1°C. During testing, the temperature ranged from 14.2 to 14.6°C.

o The report did not state how the test temperature was controlled and the temperature measurement during this test was made every 24 hours. The SEP states that temperature should be measured continuously (hourly) if it is controlled by air temperature and every 6 hours if it is controlled by a water bath.

o The SEP recommends a 16-hour light and an 8-hour dark photoperiod with a 15- to 30-minute transition period between light and dark. The report did not state whether a 15- to 30-minute transition period between light and dark was maintained.

B. **Statistical Analysis:** Statistical analyses was not needed since no mortality occurred during the test.

C. **Discussion/Results:** This study is scientifically sound. However, it does not fulfill the guideline requirements for an acute test using freshwater fish because the dosage level selected for the test was less than 100 mg/L but not high enough to produce any effects on the fish. Since the test material had low water solubility and precipitated over time, a flow-through test system would have been more appropriate for this study. Another concern about this test is the dilution water used. Since the total hardness (326-344 mg/L as $CaCO_3$) was much higher than the recommended range (40-48 mg/L as $CaCO_3$), it is not certain that the hardness did not affect the toxicity results.

The 96-hour LC_{50} value of DCPA for rainbow trout was determined by the author to be >4.7 mg/L, the concentration detected at 96 hours. The reviewer calculated the mean measured concentration by using a

time-weighted average of 0-, 24-, and 96-hour values to be 6.6 mg/L. Therefore, the 96-hour LC_{50} value, based on the time-weighted mean measured concentration was determined to be >6.6 mg/L.

D. **Adequacy of the Study:**

(1) **Classification:** Supplemental.

(2) **Rationale:** The only dosage level tested was <100 mg/L but not high enough to produce any effects on the fish.

(3) **Repairability:** None.

15. **COMPLETION OF ONE-LINER:** Yes, 07-25-90.

Results:

Concentrations of T-170-1 found in test media

Nominal concentration T-170-1 (mg/l)	Measured concentration of T-170-1 (mg/l)		
	0 hrs	24 hrs	96 hrs
Solvent Control	<0.27	<0.27	<0.27
10 (Sample 1)	9.95	7.19	4.67
10 (Sample 2)	9.98	7.45	5.15
10 (Sample 3)	10.05	7.08	4.32
10 (Samples 1, 2 and 3) Mean	9.99	7.24	4.71

Note: 0.27 mg/l was the limit of detection

Validation data

Fortification level T-170-1 (mg/l)	T-181-1 found (mg/l)	Recovery factor
10.3	9.513	0.924
10.3	9.363	0.909
5.14	4.538	0.883
1.03	0.950	0.922
Mean	-	0.910 ≠
10.3*	9.290	0.902
5.14*	4.645	0.904

≠ Analytical aquaria data corrected for recovery factor (0.910)

* Additional recovery tests assayed together with aquaria samples

TABLE 1 DSK/96C

Cumulative mortality data for rainbow trout exposed for 96 hours to T-170-1

Concentration mg/l	Rep	Cumulative mortality (initial population: 10)					
		3h	6h	24h	48h	72h	96h
Control	1	0	0	0	0	0	0
	2	0	0	0	0	0	0
	3	0	0	0	0	0	0
Solvent control	1	0	0	0	0	0	0
	2	0	0	0	0	0	0
	3	0	0	0	0	0	0
10	1	0	0	0	0	0	0
	2	0	0	0	0	0	0
	3	0	0	0	0	0	0

Rep 1, 2, 3 - Replicate 1, 2 and 3

Shaughnessy No. 078701 Chemical Name DCPA Chemical Class ____ Page ____ of ____

Study/Species/Lab/ Accession	Chemical % a.i.	Results	Reviewer/ Date	Validation Status
14-Day Single Dose Oral LD_{50}		LD50 = mg/kg (95% C.L.) Contr. Mort.(%)=		
Species	____	Slope= # Animals/Level= Age(Days)= Sex =	____	____
Lab		14-Day Dose Level mg/kg/(% Mortality) ( ), ( ), ( ), ( ), ( )		
Acc.		Comments:		
14-Day Single Dose Oral LD_{50}		LD50 = mg/kg. (95% C.L.) Contr. Mort.(%)=		
Species	____	Slope= # Animals/Level= Age(Days)= Sex =	____	____
Lab		14-Day Dose Level mg/kg/(% Mortality) ( ), ( ), ( ), ( ), ( )		
Acc.		Comments:		
8-Day Dietary LC_{50}		LC50 = ppm (95% C.L.) Contr. Mort.(%)=		
Species		Slope= # Animals/Level= Age(Days)= Sex =		
Lab	____	8-Day Dose Level ppm/(%Mortality) ( ), ( ), ( ), ( ), ( )	____	____
Acc.		Comments:		
8-Day Dietary LC_{50}		LC50 = ppm (95% C.L.) Contr. Mort.(%)=		
Species		Slope= # Animals/Level= Age(Days)= Sex =		
Lab	____	8-Day Dose Level ppm/(%Mortality) ( ), ( ), ( ), ( ), ( )	____	____
Acc.		Comments:		
48-Hour LC_{50}		LC50 = pp__ (95% C.L.) Contr. Mort.(%)= Sol. Contr. Mort.(%)=		
Species		Slope= # Animals/Level= Temperature =		
Lab	____	48-Hour Dose Level pp /(%Mortality) ( ), ( ), ( ), ( ), ( )	____	____
Acc.		Comments:		
96-Hour LC_{50}		LC50 = >6.6 pp m (95% C.L. N/A) Con. Mort(%)= 0 Sol. Con. Mor.(%)= 0	H.R. 7/25/90 Suppl	
Species Salmo gairdneri 96.7%		Slope= N/A # Animals/Level= 30 Temp.= 14.2-14.8		
Lab Huntingdon Research Centre		96-Hour Dose Level ppm/(%Mortality) 6.6 (0), ( ), ( ), ( ), ( )		
MRID # 410548-26		Comments: Based on the weighted mean measured concentration		
96-Hour LC_{50}		LC50 = pp__ (95% C.L.) Con. Mort.(%)= Sol. Con. Mort.(%)=		
Species		Slope= # Animals/Level= Temp.=		
Lab	____	96-Hour Dose Level pp /(%Mortality) ( ), ( ), ( ), ( ), ( )	____	____
Acc.		Comments:		